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FORM PTO-1390 (Modified) (REV 11-2000) U.S. DEPARTMENT OF COMMERCE PATENT AND TRADEMARK OFFICE

TRANSMITTAL LETTER TO THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US) CONCERNING A FILING UNDER 35 U.S.C. 371

ATTORNEY'S DOCKET NUMBER: 100725- /Kreisler 1099-KGB

U.S. APPLICATION NO. (IF KNOWN, SEE 37 CFR): 09/980058

INTERNATIONAL APPLICATION NO.	INTERNATIONAL FILING DATE	PRIORITY DATE CLAIMED
PCT/DE00/01854	2. Juni. 2000 (02.06.00)	4. Juni 1999 (04.06.99)

TITLE OF INVENTION
PEPTIDES FOR VACCINATION AGAINST HUMAN CMV

APPLICANT(S) FOR DO/EO/US
Florian KERN, Hans-Dieter VOLK, Petra REINKE, Nicole FAULHABER, Ingolf-Pascal SUREL, and Elham KHATAMZAS

Applicant herewith submits to the United States Designated/Elected Office (DO/EO/US) the following items and other information:

1. ☒ This is a **FIRST** submission of items concerning a filing under 35 U.S.C. 371.
2. ☐ This is a **SECOND** or **SUBSEQUENT** submission of items concerning a filing under 35 U.S.C. 371.
3. ☒ This is an express request to begin national examination procedures (35 U.S.C. 371(f)). The submission must include items (5), (6), (9) and (24) indicated below.
4. ☒ The US has been elected by the expiration of 19 months from the priority date (Article 31).
5. ☒ A copy of the International Application as filed (35 U.S.C. 371 (c) (2))
   a. ☐ is attached hereto (required only if not communicated by the International Bureau).
   b. ☒ has been communicated by the International Bureau.
   c. ☐ is not required, as the application was filed in the United States Receiving Office (RO/US).
6. ☒ An English language translation of the International Application as filed (35 U.S.C. 371(c)(2)).
   a. ☒ is attached hereto.
   b. ☐ has been previously submitted under 35 U.S.C. 154(d)(4).
7. ☒ Amendments to the claims of the International Application under PCT Article 19 (35 U.S.C. 371 (c)(3))
   a. ☐ are attached hereto (required only if not communicated by the International Bureau).
   b. ☒ have been communicated by the International Bureau.
   c. ☐ have not been made; however, the time limit for making such amendments has NOT expired.
   d. ☐ have not been made and will not be made.
8. ☒ An English language translation of the amendments to the claims under PCT Article 19 (35 U.S.C. 371(c)(3)).
9. ☐ An oath or declaration of the inventor(s) (35 U.S.C. 371 (c)(4)).
10. ☐ An English language translation of the annexes to the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371 (c)(5)).
11. ☒ A copy of the International Preliminary Examination Report (PCT/IPEA/409).
12. ☐ A copy of the International Search Report (PCT/ISA/210).

Items 13 to 20 below concern document(s) or information included:

13. ☐ An Information Disclosure Statement under 37 CFR 1.97 and 1.98.
14. ☐ An assignment document for recording. A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included.
15. ☒ A **FIRST** preliminary amendment.
16. ☐ A **SECOND** or **SUBSEQUENT** preliminary amendment.
17. ☐ A substitute specification.
18. ☐ A change of power of attorney and/or address letter.
19. ☒ A computer-readable form of the sequence listing in accordance with PCT Rule 13ter.2 and 35 U.S.C. 1.821 - 1.825.
20. ☒ A second copy of the published international application under 35 U.S.C. 154(d)(4).
21. ☐ A second copy of the English language translation of the international application under 35 U.S.C. 154(d)(4).
22. ☒ Certificate of Mailing by Express Mail
23. ☐ Other items or information.
    - Statement Regarding Sequence Listing

U.S. APPLICATION NO. (IF KNOWN, SEE 37 CFR)	INTERNATIONAL APPLICATION NO	ATTORNEY'S DOCKET NUMBER
09/980058	PCT/DE00/01854	100725- /Kreisler

24. The following fees are submitted:.

BASIC NATIONAL FEE (37 CFR 1.492 (a) (1) - (5)) :

		CALCULATIONS PTO USE ONLY
☐	Neither international preliminary examination fee (37 CFR 1.482) nor international search fee (37 CFR 1.445(a)(2)) paid to USPTO and International Search Report not prepared by the EPO or JPO	**$1040.00**
☒	International preliminary examination fee (37 CFR 1.482) not paid to USPTO but International Search Report prepared by the EPO or JPO	**$890.00**
☐	International preliminary examination fee (37 CFR 1.482) not paid to USPTO but international search fee (37 CFR 1.445(a)(2)) paid to USPTO	**$740.00**
☐	International preliminary examination fee (37 CFR 1.482) paid to USPTO but all claims did not satisfy provisions of PCT Article 33(1)-(4)	**$710.00**
☐	International preliminary examination fee (37 CFR 1.482) paid to USPTO and all claims satisfied provisions of PCT Article 33(1)-(4)	**$100.00**

ENTER APPROPRIATE BASIC FEE AMOUNT =	**$890.00**	
Surcharge of **$130.00** for furnishing the oath or declaration later than ☐ 20 ☐ 30 months from the earliest claimed priority date (37 CFR 1.492 (e)).	**$0.00**	

CLAIMS	NUMBER FILED	NUMBER EXTRA	RATE		
Total claims	16 - 20 =	0	x $18.00	**$0.00**	
Independent claims	2 - 3 =	0	x $84.00	**$0.00**	
Multiple Dependent Claims **(check if applicable)**.			☐	**$0.00**	
TOTAL OF ABOVE CALCULATIONS =				**$890.00**	
☐ Applicant claims small entity status. See 37 CFR 1.27). The fees indicated above are reduced by 1/2.				**$0.00**	
SUBTOTAL =				**$890.00**	
Processing fee of **$130.00** for furnishing the English translation later than ☐ 20 ☐ 30 months from the earliest claimed priority date (37 CFR 1.492 (f)). +				**$0.00**	
TOTAL NATIONAL FEE =				**$890.00**	
Fee for recording the enclosed assignment (37 CFR 1.21(h)). The assignment must be accompanied by an appropriate cover sheet (37 CFR 3.28, 3.31) **(check if applicable).** ☐				**$0.00**	
TOTAL FEES ENCLOSED =				**$890.00**	
				Amount to be: refunded	$
				charged	$

a. ☐ A check in the amount of _____ to cover the above fees is enclosed.

b. ☒ Please charge my Deposit Account No. **14-1263** in the amount of **$890.00** to cover the above fees. A duplicate copy of this sheet is enclosed.

c. ☒ The Commissioner is hereby authorized to charge any additional fees which may be required, or credit any overpayment to Deposit Account No. **14-1263** A duplicate copy of this sheet is enclosed.

d. ☐ Fees are to be charged to a credit card. **WARNING:** Information on this form may become public. **Credit card information should not be included on this form.** Provide credit card information and authorization on PTO-2038.

NOTE: Where an appropriate time limit under 37 CFR 1.494 or 1.495 has not been met, a petition to revive (37 CFR 1.137(a) or (b)) must be filed and granted to restore the application to pending status.

SEND ALL CORRESPONDENCE TO:

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Kurt G. Briscoe
NAME

33,141
REGISTRATION NUMBER

11-28-01
DATE

Express Mail Label No. EV 015940605 US
Date Mailed: November 28, 2001

Kreisler 1099-KGB
012823us/JH/ml

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

APPLICANTS	:	FLORAN KERN, et al
SERIAL NO.	:	TO BE ASSIGNED
FILED	:	HEREWITH
FOR	:	PEPTIDES FOR VACCINATION AGAINST HUMAN CMV
ART UNIT	:	TO BE ASSIGNED
EXAMINER	:	TO BE ASSIGNED

November 28, 2001

Hon. Commissioner of Patents
Washington, D.C. 20231

PRELIMINARY AMENDMENT

SIR:

Prior to examination, please amend the above-identified application as follows:

IN THE SPECIFICATION:

Insert as the first paragraph of the specification the following new paragraph: -- This application is a 371 of PCT/DE00/01854 filed on June 2, 2000.--

IN THE CLAIMS:

Please amend claims 7, 10-14 and 16 as follows:

7. The peptides or peptide derivatives according to claim 1, wherein R_N represents -H or an amino protective group and R_C represents -OH or a carboxy protective group.

10. The peptides or peptide derivatives according to claim 1 as a medicament or diagnostic agent.

11. Method of using a peptide or peptide derivative according to claim 1 for vaccination against HCMV infections.

12. Method of using a peptide or peptide derivative according to claim 1 as a diagnostic agent for identifying a response of the cellular immune system against HCMV.

13. Method of using a peptide or peptide derivative according to claim 1 as a diagnostic agent for quantifying a response of the cellular immune system against HCMV.

14. DNA which codes for one of the amino acid sequences and their derivatives according to claim 1.

16. A medicament comprising a DNA according to claim 14 or a plasmid or vector comprising said DNA.

REMARKS

Applicants intend to begin the national stage examination based on the amended claims filed on August 6, 2001.

The amendments above remove multiple dependencies to reduce costs, convert the use claims to the more conventional method of use format, and otherwise place the claims in better form for U.S. examination.

Early and favorable action is earnestly solicited.

Respectfully submitted,

NORRIS McLAUGHLIN & MARCUS, P.A.

By _____
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MARK-UP SHOWING THE CHANGES MADE IN THE PREVIOUS CLAIM TO YIELD THE CLAIM AS AMENDED ABOVE

7. The peptides or peptide derivatives according to [any of the preceding claims] **claim 1**, wherein R_N represents -H or an amino protective group and R_C represents -OH or a carboxy protective group.

10. The peptides or peptide derivatives according to [any of the preceding claims] **claim 1** as a medicament or diagnostic agent.

11. [Use of] **Method of using** a peptide or peptide derivative according to [any of the preceding claims for preparing a medicament] **claim 1** for vaccination against HCMV infections.

12. [Use of] **Method of using** a peptide or peptide derivative according to [any of claims 1 to 9 for preparing] **claim 1 as** a diagnostic agent for identifying a response of the cellular immune system against HCMV.

13. [Use of] **Method of using** a peptide or peptide derivative according to [any of claims 1 to 9 for preparing] **claim 1 as** a diagnostic agent for quantifying a response of the cellular immune system against HCMV.

14. DNA which codes for one of the amino acid sequences and their derivatives according to [any of claims 1 to 9] **claim 1**.

16. A medicament comprising a DNA[, plasmid or vector] according to claim 14 or [15] **a plasmid or vector comprising said DNA**.

CLAIMS:

(amended August 6, 2001)

1. Peptides having the sequence (I)

R_N - Ala Arg Ala Lys Lys Asp Glu Leu Arg Arg Lys Met Met Tyr Met- R_C (I)

or peptide derivatives thereof, wherein

R_N represents -H or an amino protective group, or at least one further amino acid outside the peptide or peptide derivative;

R_C represents -OH or a carboxy protective group, or at least one further amino acid outside the peptide or peptide derivative;

wherein said peptide derivatives have a deletion, insertion or substitution of one, two or three amino acids of sequence (I), or sequence (I) is truncated to nine contiguous amino acids, the deletion being an N-terminal and/or C-terminal deletion; and

wherein said peptide derivatives essentially have the functionality of the peptide of sequence (I) or of one of the following peptides:

Glu Leu Arg Arg Lys Met Met Tyr Met
Asp Glu Leu Arg Arg Lys Met Met Tyr
Asp Glu Leu Arg Arg Lys Met Met Tyr Met or
Asp Glu Leu Arg Arg Lys Met Met Tyr Met

(each of the above sequences = reference sequence);
i.e., to induce the production of interferon-γ or TNF-α in $CD8^+$ T cells, especially from subjects immunized with HCMV and having the appropriate HLA type.

2. The peptides or peptide derivatives according to claim 1 having the sequence

R_N - Ala Arg Ala Lys Lys Asp Glu Leu Arg Arg Lys Met Met Tyr Met- R_C
R_N - Asp Glu Leu Arg Arg Lys Met Met Tyr Met- R_C
R_N - Glu Leu Arg Arg Lys Met Met Tyr Met- R_C
R_N – Asp Glu Leu Arg Arg Lys Met Met Tyr - R_C
R_N – Asp Glu Leu Arg Arg Lys Met Met Tyr Met - R_C

3. The peptides or peptide derivatives according to claim 1, wherein said fragments are nonamers formed by truncating sequence (I) to nine contiguous amino acids, wherein the deletion is an N-terminal and/or C-terminal deletion and wherein the functionality of at least one peptide from the group of reference sequences is essentially met by said nonamer.

4. Peptides having the sequence (II)

R_N - Glu Phe Cys Arg Val Leu Cys Cys Tyr Val Leu Glu Glu Thr Ser- R_C (II)

or peptide derivatives thereof, wherein

R_N represents -H or an amino protective group, or at least one further amino acid outside the peptide or peptide derivative;

R_C represents -OH or a carboxy protective group, or at least one further amino acid outside the peptide or peptide derivative;

wherein said peptide derivatives have a deletion, insertion or substitution of one, two or three amino acids of sequence (II), or sequence (II) is truncated to nine contiguous amino acids, the deletion being an N-terminal and/or C-terminal deletion; and

wherein said peptide derivatives essentially have the functionality of the peptide of sequence (II) or of one of the following peptides:

Cys Arg Val Leu Cys Cys Tyr Val Leu
Arg Val Leu Cys Cys Tyr Val Leu Glu
Val Leu Cys Cys Tyr Val Leu Glu Glu

(each of the above sequences = reference sequence);
i.e., to induce the production of interferon-γ or TNF-α in $CD8^+$ T cells, especially from subjects immunized with HCMV and having the appropriate HLA type.

5. The peptides or peptide derivatives according to claim 1 having the sequence

R_N - Glu Phe Cys Arg Val Leu Cys Cys Tyr Val Leu Glu Glu Thr Ser- R_C
R_N - Cys Arg Val Leu Cys Cys Tyr Val Leu - R_C
R_N - Arg Val Leu Cys Cys Tyr Val Leu Glu - R_C
R_N - Val Leu Cys Cys Tyr Val Leu Glu Glu - R_C

6. The peptides or peptide derivatives according to claim 4, wherein said fragments are nonamers formed by truncating sequence (II) to nine contiguous amino acids, wherein the deletion is an N-terminal and/or C-terminal deletion and wherein the functionality of at least one peptide from the group of reference sequences is essentially met by said nonamer.

7. The peptides or peptide derivatives according to any of the preceding claims, wherein R_N represents -H or an amino protective group and R_C represents -OH or a carboxy protective group.

8. The peptides or peptide derivatives according to claim 7, wherein R_N represents -H or an acyl group and R_C represents -OH or an amino group.

9. The peptides or peptide derivatives according to claim 8, wherein R_N represents -H and R_C represents -OH.

10. The peptides or peptide derivatives according to any of the preceding claims as a medicament or diagnostic agent.

11. Use of a peptide or peptide derivative according to any of the preceding claims for preparing a medicament for vaccination against HCMV infections.

12. Use of a peptide or peptide derivative according to any of claims 1 to 9 for preparing a diagnostic agent for identifying a response of the cellular immune system against HCMV.

13. Use of a peptide or peptide derivative according to any of claims 1 to 9 for preparing a diagnostic agent for quantifying a response of the cellular immune system against HCMV.

14. DNA which codes for one of the amino acid sequences and their derivatives according to any of claims 1 to 9.

15. Vectors or plasmids into which the DNA according to claim 14 has been incorporated.

16. A medicament comprising a DNA, plasmid or vector according to claim 14 or 15.

SMB

Peptides for Vaccination Against Human CMV

The present invention relates to peptides or peptide derivatives which are useful for vaccination against human cytomegalovirus (HCMV) or for diagnoses in patients, where it can be examined whether such patients are building or have built an immune response against HCMV.

The human cytomegaloviruses (HCMV) are a group of related viruses which belong to the herpes viruses (Lutz Schneider, 1990, Pharmazie, Vol. 135, No. 27, 2396-2400). After a primary infection, the viruses remain in the body in a latent state. Physical or psychic stress can cause reactivation of latent HCMV. The virus is ubiquitous. The contamination rate of the population is around 65% in Central Europe. The cell-mediated immune response plays an essential role in the control and defense against the HCMV infection. When HCMV-specific $CD8^+$ T cells were transferred from a donor to a patient suffering from HCMV, an immune response against the HCMV infection could be observed (P.D. Greenberg et al., 1991, Development of a treatment regimen for human cytomegalovirus (CMV) infection in bone marrow transplantation recipients by adoptive transfer of donor-derived CMV-specific T cell clones expanded in vitro. Ann. N.Y. Acad. Sci., Vol.: 636, pp 184-195). Unfortunately, only few epitopes of HCMV are known which are specifically recognized by $CD8^+$ T cells. It is assumed that the immune response is essentially dominated by the 55 kD immediate-early protein 1 (IE-1) and the 65 kD lower matrix phosphoprotein (pp65) (N.J. Alp et al., 1991, Fine specificity of cellular immune responses in humans to human cytomegalovirus immediate-early 1 protein, J. Virol., Vol: 65, pp 4812-4820; and E.H. McLaughlin - Taylor et al., 1994, Identification of the major late human cytomegalovirus matrix protein pp65 as a target antigen for $CD8^+$ virus-specific cytotoxic T lymphocytes, J. Med. Virol., Vol.: 43, pp 103-110; and further, M. Wills et al., 1996, The human

cytotoxic T-lymphocyte (CTL) response to cytomegalovirus is dominated by structural protein pp65: frequency, specificity, and T-cell receptor usage of pp65-specific CTL, J. Virol. 1996, Vol. 70, pp 7569-79).

In adults having a functional immune system, the infection has an uneventful course, at most showing non-specific symptoms, such as exhaustion and slightly increased body temperature. In immunodeficient adults, pulmonary diseases and retinitis are prevailing after HCMV infections. In AIDS patients, CMV infections are the cause of numerous deaths.

Various substances are employed for treatment against cytomegalovirus. For example, Foscarnet is an antiviral substance which exhibits selective activity, as established in cell cultures, against human herpes viruses, such as herpes simplex, varicella zoster, Epstein-Barr and cytomegaloviruses, as well as hepatitis viruses. The antiviral activity is based on the inhibition of viral enzymes, such as DNA polymerases and reverse transcriptases. On cytomegaloviruses, Foscarnet has a virostatic effect, but the viruses cannot be eliminated (Lutz Schneider, 1991, Neue Arzneistoffe, Vol 136, No. 46). Another problem encountered in cytomegalovirus infections is the necessity of a permanent, sometimes life-long, treatment of the patients (especially in AIDS). A further disadvantage is the fact that the cytomegaloviruses have become more resistant against the usually employed antiviral substances in recent years (e.g., Stanat et al., 1991, Antimicrob. Agents, Chemother. Vol 35, No. 11: 2191-2197 and Knox et al., 1991, Lancet, Vol 337: 1292-1293).

The sequence of 55 kD immediate-early protein 1 (IE-1) is described and defined in A. Akrigg, G.W.G. Wilkinson, J.D. Oram (1985), The structure of the major immediate early gene of human cytomegalovirus strain AD169, Virus Res., 2:107-121. The sequence of 55 kD immediate-early protein 1 has been deposited in the Swiss-Prot Data Base, European Bioinformatics Institute, under the number P13202 (= primary accession number). The 65 kD lower matrix phosphoprotein (pp65) is described in B. Rueger, S. Klages, B. Walla et al. (1987), Primary structure and transcription of the genes coding for the two virion phosphoproteins pp65 and pp71 of human cytomegalovirus, J. Virol. 61:446-

453. The sequence of 65 kD lower matrix phosphoprotein has been deposited in the Swiss-Prot Data Base, European Bioinformatics Institute, under the number P06725 (= primary accession number). The sequences of both proteins are described in M.S. Chee, A.T. Bankier, S. Becks et al. (1990), Analysis of the protein-coding content of the sequence of human cytomegalovirus strain AD169, Curr. Top. Mircrobiol. Immunol. 154:125-169.

It has been the object of the invention to provide peptides or derivatives thereof which induce the production of interferon-γ or tumor necrosis factor α (TNF-α) in $CD8^+$ T cells, especially from subjects immunized with HCMV and having the appropriate HLA type. These peptides and their derivatives are suitable as agents for vaccination. Also, they are suitable for diagnoses to be able to establish whether a cellular immune response directed against HCMV exists in a subject, and to quantify it.

This object is achieved by peptides or peptide derivatives thereof selected from the following group of sequences:

R_N – Gln Thr Met Leu Arg Lys Glu Val Asn Ser Gln Leu Ser Leu Gly - R_C
R_N – Cys Asn Glu Asn Pro Glu Lys Asp Val Leu Ala Glu Leu Val Lys - R_C
R_N – Leu Val Lys Gln Ile Lys Val Arg Val Asp Met Val Arg His Arg- R_C
R_N – Ala Ala Asn Lys Leu Gly Gly Ala Leu Gln Ala Lys Ala Arg Ala - R_C
R_N - Ala Arg Ala Lys Lys Asp Glu Leu Arg Arg Lys Met Met Tyr Met- R_C
R_N - Asp Glu Leu Arg Arg Lys Met Met Tyr Met- R_C
R_N - Glu Leu Arg Arg Lys Met Met Tyr Met Cys Tyr Arg Asn Ile Glu- R_C
R_N – Val Thr Ser Asp Ala Cys Met Met Thr Met Tyr Gly Gly Ile Ser- R_C
R_N - Glu Phe Cys Arg Val Leu Cys Cys Tyr Val Leu Glu Glu Thr Ser- R_C
R_N – Met Ser Ile Tyr Val Tyr Ala Leu Pro Leu Lys Met Leu Asn Ile- R_C
R_N – Val Tyr Ala Leu Pro Leu Lys Met Leu Asn Ile Pro Ser Ile Asn - R_C
R_N – Ala Leu Pro Leu Lys Met Leu Asn Ile - R_C
R_N – His Ile Met Leu Asp Val Ala Phe Thr Ser His Glu His Phe Gly - R_C
R_N – Asp Val Ala Phe Thr Ser His Glu His Phe Gly Leu Leu Cys Pro- R_C
R_N – Val Ala Phe Thr Ser His Glu His Phe- R_C
R_N – Ala Phe Thr Ser His Glu His Phe Gly - R_C

R_N – Ala Asn Asp Ile Tyr Arg Ile Phe Ala Glu Leu Glu Gly Val Trp- R_C

R_N – Val Cys Ser Met Glu Asn Thr Arg Ala Thr Lys Met Gln Val Ile- R_C

R_N – Glu Asn Thr Arg Ala Thr Lys Met Gln Val Ile Gly Asp Gln Tyr- R_C

R_N – Asn Thr Arg Ala Thr Lys Met Gln Val- R_C

R_N – Thr Arg Ala Thr Lys Met Gln Val Ile - R_C

R_N – Gln Pro Phe Met Arg Pro His Glu Arg Asn Gly Phe Thr Val Leu - R_C

R_N – Pro Leu Lys Met Leu Asn Ile Pro Ser Ile Asn Val His His Tyr- R_C

R_N – Leu Asn Ile Pro Ser Ile Asn Val His His Tyr Pro Ser Ala Ala - R_C

R_N – Glu Asp Val Pro Ser Glu Lys Leu Phe Met His Val Thr Leu Gly - R_C

R_N - Cys Arg Val Leu Cys Cys Tyr Val Leu - R_C

R_N - Arg Val Leu Cys Cys Tyr Val Leu Glu - R_C

R_N - Val Leu Cys Cys Tyr Val Leu Glu Glu - R_C

R_N - Glu Leu Arg Arg Lys Met Met Tyr Met- R_C

R_N – Asp Glu Leu Arg Arg Lys Met Met Tyr - R_C

R_N – Asp Glu Leu Arg Arg Lys Met Met Tyr Met - R_C

R_N – Asp Glu Glu Glu Ala Ile Val Ala Tyr Tyr Leu Ala Thr Ala Gly - R_C

or

R_N – Glu Asn Ser Asp Gln Glu Glu Ser Glu Gln Ser Asp Glu Glu Glu - R_C

wherein

R_N represents -H or an amino protective group, or at least one further amino acid outside the peptide or peptide derivative;

R_C represents -OH or a carboxy protective group, or at least one further amino acid outside the peptide or peptide derivative;

wherein said peptide derivatives have a deletion, insertion or substitution of one, two or three amino acids of the above mentioned sequences, or the sequence is truncated to nine contiguous amino acids, the deletion being an N-terminal and/or C-terminal deletion;

wherein said peptide derivatives essentially have the functionality of one of the explicitly stated peptides:

Cys Arg Val Leu Cys Cys Tyr Val Leu
Arg Val Leu Cys Cys Tyr Val Leu Glu
Val Leu Cys Cys Tyr Val Leu Glu Glu
Glu Leu Arg Arg Lys Met Met Tyr Met
Asp Glu Leu Arg Arg Lys Met Met Tyr
Asp Glu Leu Arg Arg Lys Met Met Tyr Met
Gln Thr Met Leu Arg Lys Glu Val Asn Ser Gln Leu Ser Leu Gly
Cys Asn Glu Asn Pro Glu Lys Asp Val Leu Ala Glu Leu Val Lys
Leu Val Lys Gln Ile Lys Val Arg Val Asp Met Val Arg His Arg
Ala Ala Asn Lys Leu Gly Gly Ala Leu Gln Ala Lys Ala Arg Ala
Ala Arg Ala Lys Lys Asp Glu Leu Arg Arg Lys Met Met Tyr Met
Asp Glu Leu Arg Arg Lys Met Met Tyr Met
Glu Leu Arg Arg Lys Met Met Tyr Met Cys Tyr Arg Asn Ile Glu
Val Thr Ser Asp Ala Cys Met Met Thr Met Tyr Gly Gly Ile Ser
Glu Phe Cys Arg Val Leu Cys Cys Tyr Val Leu Glu Glu Thr Ser
Met Ser Ile Tyr Val Tyr Ala Leu Pro Leu Lys Met Leu Asn Ile
Val Tyr Ala Leu Pro Leu Lys Met Leu Asn Ile Pro Ser Ile Asn

Ala Leu Pro Leu Lys Met Leu Asn Ile
His Ile Met Leu Asp Val Ala Phe Thr Ser His Glu His Phe Gly
Asp Val Ala Phe Thr Ser His Glu His Phe Gly Leu Leu Cys Pro
Val Ala Phe Thr Ser His Glu His Phe
Ala Phe Thr Ser His Glu His Phe Gly
Ala Asn Asp Ile Tyr Arg Ile Phe Ala Glu Leu Glu Gly Val Trp
Val Cys Ser Met Glu Asn Thr Arg Ala Thr Lys Met Gln Val Ile
Glu Asn Thr Arg Ala Thr Lys Met Gln Val Ile Gly Asp Gln Tyr
Asn Thr Arg Ala Thr Lys Met Gln Val
Thr Arg Ala Thr Lys Met Gln Val Ile
Gln Pro Phe Met Arg Pro His Glu Arg Asn Gly Phe Thr Val Leu
Pro Leu Lys Met Leu Asn Ile Pro Ser Ile Asn Val His His Tyr
Leu Asn Ile Pro Ser Ile Asn Val His His Tyr Pro Ser Ala Ala
Glu Asp Val Pro Ser Glu Lys Leu Phe Met His Val Thr Leu Gly
Asp Glu Glu Glu Ala Ile Val Ala Tyr Tyr Leu Ala Thr Ala Gly

or

Glu Asn Ser Asp Gln Glu Glu Ser Glu Gln Ser Asp Glu Glu Glu

(each of the above sequences = reference sequence);
i.e., to induce the production of interferon-γ or TNF-α in $CD8^+$ T cells, especially from subjects immunized with HCMV and having the appropriate HLA type.

An appropriate HLA (human leukocyte antigen) type is a combination of MHC (major histocompatibility complex) class I molecules one or more of which enable the presentation of the peptides described. Only when an appropriate HLA type is present, a particular peptide can be presented, and only in this case, stimulation can occur. The following is a suitable reference for MHC as such and the antigen recognition of T cells: Abdul K. Abbas, Cellular and Molecular Immunology/A.K. Abbas, A.H. Lichtman, J.S. Prober, Chapters 5 and 6, 1991, W.B. Saunders, Philadelphia, ISBN 0-7216-3032-4.

Many patients which may be capable of vaccination have already been infected. Thus, in these patients, there will be enhancement of the immune response ("boosting"). The infection may be latent or actively manifest. Subjects having a latent virus are also to be considered "infected".

Preferred reference sequences are:

Val Cys Ser Met Glu Asn Thr Arg Ala Thr Lys Met Gln Val Ile;
Asn Thr Arg Ala Thr Lys Met Gln Val;
Glu Phe Cys Arg Val Leu Cys Cys Tyr Val Leu Glu Glu Thr Ser;
Cys Arg Val Leu Cys Cys Tyr Val Leu.

The individual amino acids have different preferences in the respective positions.

The comparison between the different amino acids is effected by exchanging an amino acid in a defined position while the other amino acids in the remaining positions are maintained. Generally, multiple substitutions are also possible.

The functions of the peptides or peptide derivatives are substantially changed when substituents are selected which are less conservative in substitution as

compared to the amino acids mentioned in the following. Thus, the substituents Gly and Ser are similar to the amino acid Ala, and the substituent Lys is similar to the amino acid Arg. The substituents Gln and His are similar to the amino acid Asn; the substituent Glu is similar to the amino acid Asp; the substituent Ser is similar to the amino acid Cys; the substituent Asn is similar to the amino acid Gln; the substituent Asp is similar to the amino acid Glu; the substituent Thr is similar to the amino acid Ser; the substituent Ser is similar to the amino acid Thr; the substituent Tyr is similar to the amino acid Trp; the substituents Ala and Pro are similar to the amino acid Gly; the substituents Asn and Gln are similar to the amino acid His; the substituents Leu and Val are similar to the amino acid Ile; the substituents Ile and Val are similar to the amino acid Leu; the substituents Arg, Gln and Glu are similar to the amino acid Lys; the substituents Leu, Tyr and Ile are similar to the amino acid Met; the substituents Met, Leu and Tyr are similar to the amino acid Phe; the substituents Trp and Phe are similar to the amino acid Tyr; and the substituents Ile and Leu are similar to the amino acid Val.

Such substantial changes can be achieved by substitutions with amino acids which are more different in their structures and functional groups. The effects of substantial changes are a significant change of the three-dimensional structure and/or, for example, an influence on the sheet structure or helical structure. Interactions between charges and hydrophobic chains are also to be observed in the changes.

Such analyses through substitutions can be easily accomplished. Thus, one amino acid in one position at a time is exchanged with, preferably, alanine or another amino acid. After the synthesis of the modified protein, the function of the altered protein is measured. The functions and their measurement are illustrated in the Examples. Multiple substitutions are also possible.

Definitions

The abbreviations used in the text are determined by rules which have been established by the IUPAC-IUB Commission for biochemical nomenclature (Biochemistry 11: 1726 (1972), and Biochem. J. 219: 345 (1984)). The following usual abbreviations are used: Ala = A = alanine; Arg = R = arginine; Asn = N =

asparagine; Asp = D = aspartic acid; Cys = C = cysteine; Gln = Q = glutamine; Glu = E = glutamic acid; Gly = G = glycine; His = H = histidine; Ile = I = isoleucine; Leu = L = leucine; Lys = K = lysine; Met = M = methionine; Phe = F = phenylalanine; Pro = P = proline; Ser = S = serine; Thr = T = threonine; Trp = W = tryptophan; Tyr = Y = tyrosine; and Val = V = valine.

The protective group of residue R_N can consist of:
Alkyl, aryl, alkylaryl, aralkyl, alkylcarbonyl or arylcarbonyl groups having from 1 to 10 carbon atoms, preferably naphthoyl, naphthylacetyl, naphthylpropionyl, benzoyl groups, or an acyl group having from 1 to 7 carbon atoms.

The protective group of residue R_C can consist of:
An alkoxy or aryloxy group having from 1 to 10 carbon atoms, or an amino group.

Further protective groups, for both R_N and R_C, are described in Houben-Weyl (1974), Georg Thieme Verlag, 4th Edition. The description of the protective groups in the stated literature is included herein by reference.

The sequences of the peptides or peptide derivatives according to the invention can be connected with further flanking amino acid sequences instead of a protective group on the N-terminal and/or C-terminal ends. These further flanking amino acid sequences are not essential to the function of the peptides or peptide derivatives according to the invention, but they may be carriers of other functions, for example, comprise enzymatic functions. Such flanking amino acid sequences are occurring in the nature. They may be, for example, the sequences of the variable region of an antibody which are positioned between the hypervariable regions. These sequences are referred to as framework sequences. Further known flanking amino acid sequences include uncleaved signal sequences of a secreted eukaryotic protein, the protein being expressed in a bacterium. Such signal sequences sometimes have no influence on the function of the subsequent protein. It is also possible to couple peptides or peptide derivatives according to the invention in succession, with flanking amino acid sequences being provided between the individual sequences. Also, fusion proteins are known in which the peptide is linked

through a peptide bond on the N-terminal or C-terminal end. Such a fusion protein can be expressed by bacteria or eukaryotic cells.

In particular cases, in order to decide whether a particular peptide or peptide derivative according to the invention having at least one flanking amino acid sequence and/or at least one protective group belongs to the subject matter of the invention, a comparison is to be made between:

(i) this peptide or peptide derivative with the flanking amino acid sequence and/or with the protective group; and

(ii) the same peptide or peptide derivative without the flanking amino acid sequence and without the protective group.

Both molecules should essentially have the same function as the peptides from the group of reference sequences, i.e., to induce the production of interferon-γ or TNF-α in $CD8^+$ T cells, especially from subjects immunized with HCMV and having the appropriate HLA type.

The stated amino acids are natural or artificial amino acids. The artificial amino acids are described in Houben-Weyl (1974), Georg Thieme Verlag, 4th Edition. The exchange of a described amino acid with another amino acid which belongs to the group of natural or artificial amino acids is easy to perform. Based on the test system and the comparison with one of the peptides from the group of reference sequences explicitly stated above, it is easy to find out whether an effect exists which is equal or similar to the effect of the found substances according to the invention. All D-amino acids and all amino acids which can be prepared synthetically (Houben-Weyl) also fall under the scope of protection. For the skilled person, it is easy to modify the molecular structure while retaining the essential components so that the functions which can be checked in the test are maintained. Such functionally equivalent molecules also fall under the concept of peptide derivatives.

Advantages

The peptides or peptide derivatives according to the invention allow to selectively produce vaccines against infection by human cytomegalovirus. The peptides and peptide derivatives may also be used as diagnostic agents for determining whether the subjects to be examined, especially those having the appropriate HLA type, possess $CD8^+$ T cells which are induced by substances according to the invention to production of interferon-γ or TNF-α (tumor necrosis factor α).

Preferred embodiments

Preferred sequences include nonamers formed by truncating a longer sequence as explicitly stated above or their derivatives to nine contiguous amino acids. The deletion may be an N-terminal and/or C-terminal deletion. It is essential that the functionality of a peptide from the group of reference sequences is essentially met. The reason why nonamers are very potent stimulants of $CD8^+$ T cells is the fact that MHC class I presented peptides typically have a length of nine amino acids (K.O. Falk et al. (1991), Allele-specific motifs revealed by sequencing of self-peptides eluted from MHC molecules, Nature, Vol. 351, pp 290-296, and H.G. Rammensee et al. (1999), An Internet Database for MHC Ligands and Peptide Motifs, http://134.296.221/scripts/hlaserver.dll/home.htm).

It is also possible to link the above mentioned nonamers with further amino acids through peptide bonds to form sequences of at least 10 amino acids. These sequences also have the functionality to induce the production of interferon-γ or TNF-α in $CD8^+$ T cells. Thus, the scope of protection of the invention also encompasses sequences extended by at least one amino acid of nonamers which have the functionality to induce the production of interferon-γ or TNF-α in $CD8^+$ T cells, especially from subjects immunized with HCMV and having the appropriate HLA type. For the extended nonamers, it is essential also to have the functionality to induce the production of interferon-γ or TNF-α in $CD8^+$ T cells, especially from subjects immunized with HCMV and having the appropriate HLA type.

Preparation of the peptides

Further, the peptides or peptide derivatives according to the invention can be easily prepared. Such short peptides or peptide derivatives can be prepared using a technique which is known to those skilled in the art of peptide synthesis. A survey of many of these techniques can be looked up in J.M. Stewart and J.D. Young, San Francisco, 1969; and J. Meierhofer, Hormonal Proteins and Peptides, Vol. 2, p 46, Academic Press (New York), 1973, for the solid-phase method, and E. Schroder and K. Lubke, The Peptides, Vol. 1, Academic Press (New York), 1965, for the liquid-phase method. The steps of the synthesis are described in EP-A 0 097 031. The general process steps from the European publications can be transferred by analogy to the synthesis of the peptides or peptide derivatives according to the invention as herein described. Further references relating to solid-phase synthesis include: Solid Phase Synthesis, E. Atherton and R.C. Sheppard (1989), IRL Press, ISBN 1-85221-133-4, and Amino Acid and Peptide Synthesis, J. Jones, Oxford Science Publication (1992), ISBN 0-19-855668-3.

Further embodiments in terms of the residues

In addition to modification of the amino acid sequence of the peptides or peptide derivatives according to the invention, it is also possible to vary the residues R_N and R_C. However, the residues need not influence the function. Nevertheless, parameters such as stability, pH dependence, biodegradability and interactions with the native part of the fusion protein can be significantly affected by protective groups.

Preferred are peptides or peptide derivatives according to the invention in which residue

R_N represents -H or an amino protective group; and

R_C represents -OH or a carboxy protective group.

More preferred are peptides or peptide derivatives according to the invention in which residue

R_N represents -H or an acyl group; and

R_C represents -OH or an amino group.

Most preferred are peptides or peptide derivatives according to the invention in which residue

R_N represents -H; and

R_C represents -OH.

Preparation methods for the peptides according to the invention

The invention further comprises the preparation of the peptides or peptide derivatives according to the invention, wherein an N-α-protected ω-amino-α-amino acid is reacted with a dialdehyde in the presence of a reductant, followed by deprotecting the side chains and optionally deprotecting the N-terminus and/or the C-terminus.

The invention also relates to a process in which the peptides or peptide derivatives according to the invention are prepared by condensing the amino acids in a homogeneous phase or by the solid-phase method;

wherein the carboxy end of an amino acid to be coupled whose amino groups and optionally side chain functional groups bear a protective group reacts with the free amino end of the amino acid to be coupled or the peptide to be coupled in the presence of a condensation reagent; and

in the case of a non-terminal amino acid, the α-amino protective group of the coupled amino acid is subsequently cleaved off, and other amino acids are coupled to the peptide chain to be synthesized according to the two steps described above; or

in the case of a non-terminal amino acid, optionally, the α-amino protective group of the coupled amino acid is subsequently cleaved off; and

in the case of the solid-phase method, after the coupling of the last amino acid, the peptide or peptide derivative is cleaved from the solid phase.

Use as a medicament

The peptides or peptide derivatives according to the invention are suitable for use as a medicament or diagnostic agent.

Most preferred is the use of a peptide or peptide derivative for preparing a medicament for vaccination against the human cytomegalovirus.

It is also advantageous to use a peptide or peptide derivative according to the invention for preparing a diagnostic agent for identifying a cellular immune response against HCMV. Thus, T cells of the patient can be stimulated in vitro with the peptides and peptide derivatives according to the invention. If this is accompanied by induction of the production of interferon-γ or TNF-α in $CD8^+$ T cells, an immune response against HCMV has been detected. If such stimulation does not result in the induction of interferon-γ or TNF-α in $CD8^+$ T cells of a subject immunized with HCMV and having the appropriate HLA type, this may mean that this subject has not built a $CD8^+$ T cell response against the HCMV, or an existing $CD8^+$ T cell response is not directed against the epitope used for stimulation.

It is particularly advantageous to use a peptide or peptide derivative according to the invention for preparing a diagnostic agent for identifying an immune response against HCMV in immunodeficient subjects.

The immune response against HCMV identified through the induction of INF-γ or TNF-α in $CD8^+$ T cells can be quantified by determining the number of $CD8^+$ T cells in which induction of these cytokines has occurred. This number can be stated as an absolute (per volume of the starting material) or relative value (for example, based on all $CD8^+$ T cells). Such quantification can be effected, for example, by flow cytometry or by another suitable method.

As a medicament, it is preferred for the peptides or peptide derivatives according to the invention to form a composition with pharmacologically acceptable auxiliary agents and carriers. Such auxiliary agents and carriers are described in Remington's Pharmaceutical Science, 15th Ed., Mack Publishing Company, East Pennsylvania (1980). The compositions can be prepared by known methods.

It is advantageous to use the peptides or peptide derivatives for loading dendritic cells, which are subsequently administered to a patient as a medicament. It is more advantageous to use the peptides or peptide derivatives for loading HLA-identical or partially HLA-identical dendritic cells which are subsequently administered to a patient as a medicament.

The use of peptide-loaded dendritic cells as a vaccine is described in Brugger et al., Ann. N.Y. Acad. Sci., Vol. 872, pp 363-371.

The peptides or peptide derivatives according to the invention have pharmacological properties and can therefore be used as a pharmaceutically active substance or diagnostic agent, especially as a vaccine or diagnostic agent. The invention also relates to a medicament which contains the peptides or peptide derivatives according to the invention.

The experimental results of the in-vitro testing show that the peptides or peptide derivatives according to the invention can be used as medicaments or for medical treatment. These experimental results can be transferred without any problem from the in-vitro test system to an in-vivo system.

The invention further provides

(i) the use of peptides or peptide derivatives according to the invention (for preparing a medicament) as vaccines against infections with HCMV;

(ii) a pharmacological composition as a vaccine against infections with HCMV which, for treatment or prophylaxis, comprises the peptides or peptide de-

rivatives according to the invention and at least one pharmaceutically acceptable auxiliary agent and/or carrier.

Different doses are suitable for providing a therapeutic effect. They depend, for example, on the salts employed, on the host, on the kind of administration and on the type and severity of the conditions to be treated.

Combinations of the peptides or peptide derivatives according to the invention are also possible.

The invention further relates to DNA (deoxyribonucleic acid) which codes for one of the above mentioned amino acid sequences and their derivatives.

Such DNA can be usefully incorporated in vectors or plasmids. Such vectors are capable of intruding into human cells and start protein biosynthesis therein. In this form, the protein biosynthetic apparatus of the human can be used for synthesizing and secreting the desired peptides.

With differently coding DNA, such vectors are described in D. Salmon-Ceron et al. (1999), AIDS Res. Hum. Retroviruses, Vol. 15/7, pp 633-645, and F. Dorner et al. (1999), Ann. Med. Vol. 31/1, pp 51-60, and G. Ferrari et al. (1997), Blood, Vol. 90, pp 2406-2416, and K. Molling (1998), Z. Ärztl. Fortbild. Qualitätssich., Vol. 92, pp 681-683, and M. Giese (1998), Virus Genes, Vol. 17, pp 219-232.

Examples

Methods

Citrated blood was obtained from anti-HCMV-IgG-seropositive blood donors of a defined HLA type. A Ficoll-Paque density centrifugation was performed. The cells were washed with sterile PBS and resuspended in RPMI 1640 which contained 0.1% BSA and 2 mM glutamine. The cells were adjusted to 10^7 cells per ml. Two hundred microliters of the cell suspension and of the peptide solution (10 µg per

ml in RPMI/BSA) was filled into Cellstar® polystyrene tubes and stored in an incubator.

After one hour, 1600 µl of RPMI 1640 which contained 12.5% FCS, 50 mM glutamine and 12.5 µg per ml of brefeldin A was added. After another five hours, the cells were washed with cold PBS, resuspended in PBS with 1 mM EDTA, incubated at 37 °C for another 10 minutes, and again washed with cold PBS.

After surface labeling with monoclonal antibodies for 30 minutes at 4 °C in the dark, the cells were fixed in PBS containing 4% paraformaldehyde for 4 minutes at 37 °C, followed by washing with PBS. The cells were then permeabilized using the permeabilization solution of Becton Dickinson (Heidelberg), followed by another wash with PBS.

Subsequent to the following intracellular labeling with monoclonal antibodies against interferon-γ and/or TNF-α, the cells were again washed in PBS and analyzed with a FACScalibur® flow cytometer (Becton Dickinson) using CellQuest™ software. Unstimulated samples were employed as controls.

Results

The substances according to the invention exhibited a stimulation of the $CD8^+$ T cells. Therefore, these substances are suitable as vaccines. Further, they are suitable for use as diagnostic agents, wherein the cells which can respond to HCMV are identified. The inability or ability of a patient to respond to HCMV or an immunization with HMCV which has already taken place are established by this form of diagnose. The stimulation of the $CD8^+$ T cells was detected by the presence of intracellularly retained interferon-γ or tumor necrosis factor α (TNF-α).

Preparation of the peptides

The synthesis of peptides or peptide derivatives can be performed on a Multiple Peptide Synthesizer (MPS) AMS 422 of ABIMED (Langenfeld) (H. Gausepohl et al. (1992), Peptide Research 5/6: 315-320).

As solid supports for the peptide synthesis, hardened cellulose (Whatman 540; Catalogue No. 1540917) of Whatman (Maidstone, Great Britain) and polystyrene resin Tenta Gel SRAM (capacity 0.25 meq/g) of Rapp Polymere (Tübingen, Germany) can be used.

The solid-phase peptide synthesis is described extensively in Rudolf Volkmer-Engert, Berit Hoffmann, and Jens Schneider-Mergener (1997), Stable Attachment of the HMB-Linker to Continuous Cellulose Membranes for Parallel Solid Phase Spot Syntheses, Tetrahedron Letter, Vol. 38,6; pp 1029-1032.

CLAIMS:

1. Peptides or peptide derivatives thereof selected from the following group of sequences:

R_N – Gln Thr Met Leu Arg Lys Glu Val Asn Ser Gln Leu Ser Leu Gly - R_C
R_N – Cys Asn Glu Asn Pro Glu Lys Asp Val Leu Ala Glu Leu Val Lys - R_C
R_N – Leu Val Lys Gln Ile Lys Val Arg Val Asp Met Val Arg His Arg- R_C
R_N – Ala Ala Asn Lys Leu Gly Gly Ala Leu Gln Ala Lys Ala Arg Ala - R_C
R_N - Ala Arg Ala Lys Lys Asp Glu Leu Arg Arg Lys Met Met Tyr Met- R_C
R_N - Asp Glu Leu Arg Arg Lys Met Met Tyr Met- R_C
R_N - Glu Leu Arg Arg Lys Met Met Tyr Met Cys Tyr Arg Asn Ile Glu- R_C
R_N – Val Thr Ser Asp Ala Cys Met Met Thr Met Tyr Gly Gly Ile Ser- R_C
R_N - Glu Phe Cys Arg Val Leu Cys Cys Tyr Val Leu Glu Glu Thr Ser- R_C
R_N – Met Ser Ile Tyr Val Tyr Ala Leu Pro Leu Lys Met Leu Asn Ile- R_C
R_N – Val Tyr Ala Leu Pro Leu Lys Met Leu Asn Ile Pro Ser Ile Asn - R_C
R_N – Ala Leu Pro Leu Lys Met Leu Asn Ile - R_C
R_N – His Ile Met Leu Asp Val Ala Phe Thr Ser His Glu His Phe Gly - R_C
R_N – Asp Val Ala Phe Thr Ser His Glu His Phe Gly Leu Leu Cys Pro- R_C
R_N – Val Ala Phe Thr Ser His Glu His Phe- R_C
R_N – Ala Phe Thr Ser His Glu His Phe Gly - R_C
R_N – Ala Asn Asp Ile Tyr Arg Ile Phe Ala Glu Leu Glu Gly Val Trp- R_C
R_N – Val Cys Ser Met Glu Asn Thr Arg Ala Thr Lys Met Gln Val Ile- R_C
R_N – Glu Asn Thr Arg Ala Thr Lys Met Gln Val Ile Gly Asp Gln Tyr- R_C
R_N – Asn Thr Arg Ala Thr Lys Met Gln Val- R_C
R_N – Thr Arg Ala Thr Lys Met Gln Val Ile - R_C
R_N – Gln Pro Phe Met Arg Pro His Glu Arg Asn Gly Phe Thr Val Leu - R_C
R_N – Pro Leu Lys Met Leu Asn Ile Pro Ser Ile Asn Val His His Tyr- R_C
R_N – Leu Asn Ile Pro Ser Ile Asn Val His His Tyr Pro Ser Ala Ala- R_C
R_N – Glu Asp Val Pro Ser Glu Lys Leu Phe Met His Val Thr Leu Gly - R_C

R_N - Cys Arg Val Leu Cys Cys Tyr Val Leu - R_C

R_N - Arg Val Leu Cys Cys Tyr Val Leu Glu - R_C

R_N - Val Leu Cys Cys Tyr Val Leu Glu Glu - R_C

R_N - Glu Leu Arg Arg Lys Met Met Tyr Met- R_C

R_N – Asp Glu Leu Arg Arg Lys Met Met Tyr - R_C

R_N – Asp Glu Leu Arg Arg Lys Met Met Tyr Met - R

R_N – Asp Glu Glu Glu Ala Ile Val Ala Tyr Tyr Leu Ala Thr Ala Gly - R_C

or

R_N – Glu Asn Ser Asp Gln Glu Glu Ser Glu Gln Ser Asp Glu Glu Glu - R_C

wherein

R_N represents -H or an amino protective group, or at least one further amino acid outside the peptide or peptide derivative;

R_C represents -OH or a carboxy protective group, or at least one further amino acid outside the peptide or peptide derivative;

wherein said peptide derivatives have a deletion, insertion or substitution of one, two or three amino acids of the above mentioned sequences, or the sequence is truncated to nine contiguous amino acids, the deletion being an N-terminal and/or C-terminal deletion;

wherein said peptide derivatives essentially have the functionality of one of the explicitly stated peptides:

Cys Arg Val Leu Cys Cys Tyr Val Leu
Arg Val Leu Cys Cys Tyr Val Leu Glu
Val Leu Cys Cys Tyr Val Leu Glu Glu
Glu Leu Arg Arg Lys Met Met Tyr Met
Asp Glu Leu Arg Arg Lys Met Met Tyr
Asp Glu Leu Arg Arg Lys Met Met Tyr Met
Gln Thr Met Leu Arg Lys Glu Val Asn Ser Gln Leu Ser Leu Gly
Cys Asn Glu Asn Pro Glu Lys Asp Val Leu Ala Glu Leu Val Lys
Leu Val Lys Gln Ile Lys Val Arg Val Asp Met Val Arg His Arg
Ala Ala Asn Lys Leu Gly Gly Ala Leu Gln Ala Lys Ala Arg Ala
Ala Arg Ala Lys Lys Asp Glu Leu Arg Arg Lys Met Met Tyr Met
Asp Glu Leu Arg Arg Lys Met Met Tyr Met
Glu Leu Arg Arg Lys Met Met Tyr Met Cys Tyr Arg Asn Ile Glu
Val Thr Ser Asp Ala Cys Met Met Thr Met Tyr Gly Gly Ile Ser
Glu Phe Cys Arg Val Leu Cys Cys Tyr Val Leu Glu Glu Thr Ser
Met Ser Ile Tyr Val Tyr Ala Leu Pro Leu Lys Met Leu Asn Ile
Val Tyr Ala Leu Pro Leu Lys Met Leu Asn Ile Pro Ser Ile Asn
Ala Leu Pro Leu Lys Met Leu Asn Ile
His Ile Met Leu Asp Val Ala Phe Thr Ser His Glu His Phe Gly
Asp Val Ala Phe Thr Ser His Glu His Phe Gly Leu Leu Cys Pro
Val Ala Phe Thr Ser His Glu His Phe
Ala Phe Thr Ser His Glu His Phe Gly

Ala Asn Asp Ile Tyr Arg Ile Phe Ala Glu Leu Glu Gly Val Trp
Val Cys Ser Met Glu Asn Thr Arg Ala Thr Lys Met Gln Val Ile
Glu Asn Thr Arg Ala Thr Lys Met Gln Val Ile Gly Asp Gln Tyr
Asn Thr Arg Ala Thr Lys Met Gln Val
Thr Arg Ala Thr Lys Met Gln Val Ile
Gln Pro Phe Met Arg Pro His Glu Arg Asn Gly Phe Thr Val Leu
Pro Leu Lys Met Leu Asn Ile Pro Ser Ile Asn Val His His Tyr
Leu Asn Ile Pro Ser Ile Asn Val His His Tyr Pro Ser Ala Ala
Glu Asp Val Pro Ser Glu Lys Leu Phe Met His Val Thr Leu Gly
Asp Glu Glu Glu Ala Ile Val Ala Tyr Tyr Leu Ala Thr Ala Gly

or

Glu Asn Ser Asp Gln Glu Glu Ser Glu Gln Ser Asp Glu Glu Glu

(each of the above sequences = reference sequence);
i.e., to induce the production of interferon-γ or TNF-α in $CD8^+$ T cells, especially from subjects immunized with HCMV and having the appropriate HLA type.

2. Fragments of peptides or peptide derivatives according to claim 1, wherein said fragments are nonamers formed by truncating a longer sequence according to claim 1 to nine contiguous amino acids, wherein the deletion is an N-terminal and/or C-terminal deletion and wherein the functionality of at least one peptide from the group of reference sequences is essentially met by said nonamer.

3. The peptides or peptide derivatives according to any of the preceding claims, wherein
R_N represents -H or an amino protective group; and
R_C represents -OH or a carboxy protective group.

4. The peptides or peptide derivatives according to claim 3, wherein residue
R_N represents -H or an acyl group; and
R_C represents -OH or an amino group.

5. The peptides or peptide derivatives according to claim 4, wherein residue
R_N represents -H; and
R_C represents -OH.

6. The peptides or peptide derivatives according to any of the preceding claims as a medicament or diagnostic agent.

7. Use of a peptide or peptide derivative according to any of the preceding claims for preparing a medicament for vaccination against HCMV infections.

8. Use of a peptide or peptide derivative according to any of claims 1 to 6 for preparing a diagnostic agent for identifying a response of the cellular immune system against HCMV.

9. Use of a peptide or peptide derivative according to any of claims 1 to 6 for preparing a diagnostic agent for quantifying a response of the cellular immune system against HCMV.

10. DNA which codes for one of the amino acid sequences and their derivatives according to any of claims 1 to 5.

11. Vectors or plasmids into which the DNA according to claim 10 has been incorporated.

12. The DNA, plasmid or vector according to claim 10 or 11 as a medicament.

Abstract

Peptides, which may be fragments of the IE-1 or pp65 proteins, selected from the following group of sequences:

R_N – Gln Thr Met Leu Arg Lys Glu Val Asn Ser Gln Leu Ser Leu Gly - R_C

R_N – Cys Asn Glu Asn Pro Glu Lys Asp Val Leu Ala Glu Leu Val Lys - R_C

R_N – Leu Val Lys Gln Ile Lys Val Arg Val Asp Met Val Arg His Arg- R_C

R_N – Ala Ala Asn Lys Leu Gly Gly Ala Leu Gln Ala Lys Ala Arg Ala - R_C

R_N - Ala Arg Ala Lys Lys Asp Glu Leu Arg Arg Lys Met Met Tyr Met- R_C

R_N - Asp Glu Leu Arg Arg Lys Met Met Tyr Met- R_C

R_N - Glu Leu Arg Arg Lys Met Met Tyr Met Cys Tyr Arg Asn Ile Glu- R_C

R_N – Val Thr Ser Asp Ala Cys Met Met Thr Met Tyr Gly Gly Ile Ser- R_C

R_N - Glu Phe Cys Arg Val Leu Cys Cys Tyr Val Leu Glu Glu Thr Ser- R_C

R_N – Met Ser Ile Tyr Val Tyr Ala Leu Pro Leu Lys Met Leu Asn Ile- R_C

R_N – Val Tyr Ala Leu Pro Leu Lys Met Leu Asn Ile Pro Ser Ile Asn - R_C

R_N – Ala Leu Pro Leu Lys Met Leu Asn Ile - R_C

R_N – His Ile Met Leu Asp Val Ala Phe Thr Ser His Glu His Phe Gly - R_C

R_N – Asp Val Ala Phe Thr Ser His Glu His Phe Gly Leu Leu Cys Pro- R_C

R_N – Val Ala Phe Thr Ser His Glu His Phe- R_C

R_N – Ala Phe Thr Ser His Glu His Phe Gly - R_C

R_N – Ala Asn Asp Ile Tyr Arg Ile Phe Ala Glu Leu Glu Gly Val Trp- R_C

R_N – Val Cys Ser Met Glu Asn Thr Arg Ala Thr Lys Met Gln Val Ile- R_C

R_N – Glu Asn Thr Arg Ala Thr Lys Met Gln Val Ile Gly Asp Gln Tyr- R_C

R_N – Asn Thr Arg Ala Thr Lys Met Gln Val- R_C

R_N – Thr Arg Ala Thr Lys Met Gln Val Ile - R_C

R_N – Gln Pro Phe Met Arg Pro His Glu Arg Asn Gly Phe Thr Val Leu - R_C

R_N – Pro Leu Lys Met Leu Asn Ile Pro Ser Ile Asn Val His His Tyr- R_C

R_N – Leu Asn Ile Pro Ser Ile Asn Val His His Tyr Pro Ser Ala Ala- R_C

R_N – Glu Asp Val Pro Ser Glu Lys Leu Phe Met His Val Thr Leu Gly - R_C

R_N - Cys Arg Val Leu Cys Cys Tyr Val Leu - R_C

R_N - Arg Val Leu Cys Cys Tyr Val Leu Glu - R_C

R_N - Val Leu Cys Cys Tyr Val Leu Glu Glu - R_C

R_N - Glu Leu Arg Arg Lys Met Met Tyr Met- R_C

R_N – Asp Glu Leu Arg Arg Lys Met Met Tyr - R_C

R_N – Asp Glu Leu Arg Arg Lys Met Met Tyr Met - R

R_N – Asp Glu Glu Glu Ala Ile Val Ala Tyr Tyr Leu Ala Thr Ala Gly - R_C

or

R_N – Glu Asn Ser Asp Gln Glu Glu Ser Glu Gln Ser Asp Glu Glu Glu - R_C

wherein

R_N represents -H or an amino protective group;

R_C represents -OH or a carboxy protective group.

The peptides according to the invention are used for preparing a medicament for vaccination against HCMV infections, or a diagnostic agent for identifying an immune response against HCMV.

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COMBINATION DECLARATION & POWER OF ATTORNEY

As a below named inventor, I hereby declare that:

My residence, post office address and citizenship are as stated below next to my name. I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled

PEPTIDES FOR VACCINATION AGAINST HUMAN CMV

the specification of which was filed on November 28, 2001

as Application Serial No. 09/980,058 which is a 371 of PCT/DE00/01854

I hereby state that I have reviewed and understand the contents of the above identified specification, including the claims.

I acknowledge the duty to disclose information which is material to the examination of this application in accordance with Title 37, Code of Federal Regulations §1.56(a).

I hereby claim foreign priority benefits under Title 35, United States Code, §119 of any foreign application(s) for patent or inventor's certificate listed below and have also identified below any foreign application for patent or inventor's certificate having a filing date before that of the application on which priority is claimed:

Prior Foreign Application(s)			Priority Claimed
199 27 039.2 (Number)	Germany (Country)	4 June 1999 (Day/Month/Yr. Filed)	x yes ___ no
199 43 702.5 (Number)	Germany (Country)	7 September 1999 (Day/Month/Yr. Filed)	x yes ___ no

I hereby claim the benefit under Title 35, United States Code, §120 of any United States application(s) listed below and, insofar as the subject matter of each of the claims of this application is not disclosed in the prior United States application in the manner provided by the first paragraph of Title 35, United States Code, §112, I acknowledge the duty to disclose material information as defined in Title 37, Code of Federal Regulations, §1.56(a) which occurred between the filing date of the prior application and the national or PCT international filing date of this application:

(Application Serial No.)	(Filing Date)	(Status) (patented,pending,abandoned)
(Application Serial No.)	(Filing Date)	(Status) (patented,pending,abandoned)

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punished by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

POWER OF ATTORNEY: As a named inventor, I hereby appoint the following attorney(s) and/or agent(s) to prosecute this application and transact all business in the Patent and Trademark Office connected therewith:

Kurt G. Briscoe, Reg. No. 33,141; William C. Gerstenzang, Reg. No. 27,552; Lorimer P. Brooks, Reg. No. 15,155; Bruce Londa, Reg. No. 33,531; Christa Hildebrand, Reg. No. 34,953; and Howard C. Lee, Reg. No. 48,104 all of 220 East 42nd Street, 30th Floor, New York, New York 10017; William R. Robinson, Reg. No. 27,224 of 721 Route 202-206 Bridgewater, New Jersey 08807; Davy E. Zoneraich, Reg. No. 37,267, Mark A. Montana, Reg. No. 44,948 and Robert A. Hyde, Reg. No. 46,354, of 721 Route 202-206, Bridgewater, New Jersey 08807, my attorneys with full power of substitution and revocation.

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FULL NAME OF SEVENTH INVENTOR:	
INVENTOR'S SIGNATURE:	DATE:
RESIDENCE:	CITIZENSHIP:
POST OFFICE ADDRESS:	

SEQUENCE LISTING

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Leu Val Lys Gln Ile Lys Val Arg Val Asp Met Val Arg His Arg
1 5 10 15

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against HCMV

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Val Thr Ser Asp Ala Cys Met Met Thr Met Tyr Gly Gly Ile Ser
1 5 10 15

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Thr Arg Ala Thr Lys Met Gln Val Ile
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<400> 28

Gln Pro Phe Met Arg Pro His Glu Arg Asn Gly Phe Thr Val Leu
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<400> 29

Pro Leu Lys Met Leu Asn Ile Pro Ser Ile Asn Val His His Tyr
1 5 10 15

<210> 30
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<400> 30

Leu Asn Ile Pro Ser Ile Asn Val His His Tyr Pro Ser Ala Ala
1 5 10 15

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Glu Asp Val Pro Ser Glu Lys Leu Phe Met His Val Thr Leu Gly
1 5 10 15

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<400> 32

Asp Glu Glu Glu Ala Ile Val Ala Tyr Tyr Leu Ala Thr Ala Gly
1 5 10 15

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Glu Asn Ser Asp Gln Glu Glu Ser Glu Gln Ser Asp Glu Glu Glu
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